FRACTAL DIMENSION AS A CHARACTERISTIC OF BIOLOGICAL CELL AFM IMAGES

I. E. Starodubtsev
IBA Gomel Park
Gomel, BELARUS

1 Introduction

Biological cells are open, dynamic self-organizing microsystems that exchange matter, energy and information with their environment. Performing their physiological functions, the biological cells interact with other cells, vessel walls and macromolecular complexes when as a rule they are subjected to mechanical stress.

The cell surface properties including its structural and mechanical properties are important parameters of cell state and functioning. Because the change of the cell surface properties occurs during the pathological processes, the qualitative and quantitative cell surface characteristics can be markers of cell health and pathology.

Atomic force microscopy (AFM) is one of the modern methods for studying solid surface structure and properties. AFM has tremendous advantages over electron microscopy (including scanning electron microscopy), as it allows working with objects directly both on air and in various fluids. Atomic force microscopes are widely used in many fields of science and technology: biophysics, biochemistry, materials science, pharmaceutics, surface physics, electronics and others. Nowadays AFM is used in studying the biological cells and tissues as well.

AFM provides the images of topography (topography scan mode) and spatial distribution of local physical and mechanical properties (torsion scan mode) of the studied surface with nanometer resolution (Figure 1).

AFM-image of a cell surface is a set of points with three coordinates (x, y, z) that represents either a topography map (in this case x, y and z are positions of the surface points) or map of local physical and mechanical properties (in this case x, y are positions of the surface points and z is a force value in the certain point).

The dimension is an important parameter of the surface of objects. Real surfaces are characterized by fractal (fractional) dimension. There are various methods to calculate the fractal dimension: box-counting method, power spectrum method, hand and drives method and others [1]. Each method has its own features that limit its usage.

The work aims at studying the relationship between the fractal dimension and geometrical parameters of AFM images of real biological cells and model surfaces.

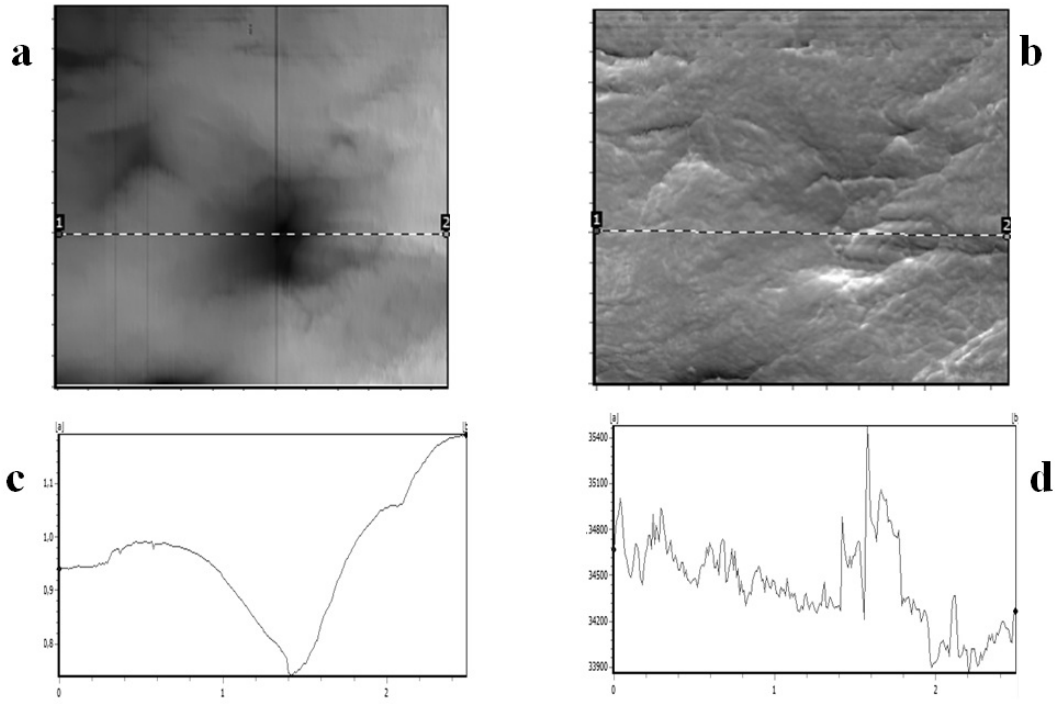

Figure 1: AFM images (a, b) and profiles (c, d) of 549 cancer cell surface (scan size is 2.5μm × 2.5μm).

2 Methods

The main method we used to calculate the fractal dimension is box-counting method [2,3]. It based on the following formula:

$$D_F = -\lim_{\varepsilon \to 0} \log_\varepsilon N(\varepsilon), \tag{1}$$

where $N(\varepsilon)$ is minimal number of cubes with edge ε that cover together the required surface.

To find the fractal dimension (D_F) the system of equations had to be solved:

$$\ln N(\varepsilon) = \ln C + D_F \ln \varepsilon, \tag{2}$$

where the number of equations is larger than the number of unknown variables. The system often has no exact solution and, therefore, is solved numerically.

In the present work, the mentioned above method was realized on C++ programming language using Borland C++ Builder IDE and STL library.

The implementation of the algorithm included the following steps. The spatial region with the studied surface was divided by the cubic lattice with cube edge ε (initially set as a half of the studied region size). Then the number of cubes $N(\varepsilon)$ that included at least one point of the surface was calculated. The cube edge ε was reduced by two and the process repeated in loop until cube edge became less than a constant

depending on AFM scanning step. At each step of loop pairs $\ln N(\varepsilon)$ and $\ln \varepsilon$ were added to resultant array. The plot $\ln N(\varepsilon)$ against $\ln \varepsilon$ was approximated with a line which slope was equal to surface fractal dimension D_F.

We used also the modified box-counting algorithm. The surface was divided into a few (from 2 to 8) equal fragments and the fractal dimensions were calculated for each fragment using box-counting algorithm. Then the fractal dimension for the whole surface was calculated using the sample of D_F and represented as the mean and confidence interval limits.

3 Results

We analyzed the change of fractal dimension with the change of the scale factor for axis Z (Z-scaling). The problem of the change of the object dimension during scaling has been recently reviewed by Simon Villerton in two-dimensional case [4]. In the present work, the analysis of the dependence of the fractal dimension on Z-scaling was performed in the following way: the data along axes X and Y were not changed but the data of axis Z was multiplied by factor t changed over a broad range of values. D_F of the whole surface was calculated for each value of factor t (scaling factor for axis Z):

$$D_F = \varphi(t). \tag{3}$$

Various modeling surfaces have been generated for the qualitative analysis of the dependencies: plane surface, plane surface with a finite number of Gaussian peaks, wave surfaces $Z = H \sin(\omega\sqrt{x^2+y^2})$ and $Z = H|\sin(\omega\sqrt{x^2+y^2})|$. The changes in $D_F = \varphi(t)$ with the changes of frequency, amplitude and other surface parameters were found and analyzed.

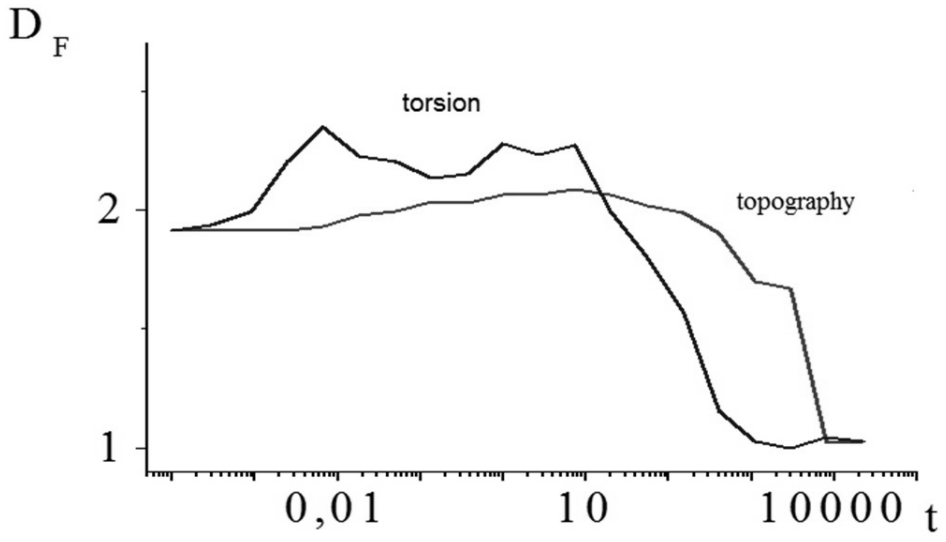

Figure 2: Dependence $D_F = \varphi(t)$ for torsion and topography scans of erythrocyte surface.

For the erythrocyte surface (Figure 2) D_F at the smaller values of t tends to 2 (plane surface) and at the larger values of t tends to 1 (line). In the intermediate range of t, function $D_F = \varphi(t)$ has some maxima. The results of the performed analysis has shown that the parameters of dependence $D_F = \varphi(t)$ was qualitatively related to the type of elements of the surface. For example, if the first peak in curve $D_F = \varphi(t)$ was higher than the second peak, the studied surface had the frequent small-scale heterogeneities, and if the second peak was higher than the first peak, the surface was relatively smooth with a few large-scale heterogeneities.

4 Conclusion

Dependence $D_F = \varphi(t)$ is a characteristic of AFM images of surfaces (including the surfaces of biological cells), which describes the surface features better than a single value D_F.

References

[1] Napolitano A., Ungania S., Cannata V. (2012). Fractal dimension estimation methods for biomedical images. *MATLAB - A fundamental tool for scientific computing and engineering applications*. Vol. **3**, pp. 161–178.

[2] Braverman B., Tambasco M. (2013). Scale-specific multifractal medical image analysis. *Computational and Mathematical Methods in Medicine*. ID 262931.

[3] Foroutan-pour K., Dutilleul P., Smith D. L. (1999). Advances in the implementation of the box-counting method of fractal dimension estimation. *Applied Mathematics and Computation*. Vol. **105**(2), pp. 195–210.

[4] Willerton S. (2015). Spread: a measure of the size of metric spaces. *Intern. J. Comput. Geom. Appl.* Vol. **25**(3), pp. 207–225.